Weed Science, 54:898–902. 2006

Factors affecting germination of horseweed (*Conyza canadensis*)

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The influence of environmental factors on germination and emergence of horseweed was examined in growth chamber experiments. Germination was highest (61%) under 24/20 C day/night temperature under light. Horseweed seed germination was observed under both light (13 h photoperiod) and complete darkness (24 h), but germination under continuous darkness was only 0 to 15% compared with 0 to 61% under light. All other experiments were conducted under 24/20 C and 13-h light conditions. Germination was 19 to 36% over a pH range from 4 to 10, with a trend toward higher germination under neutral-to-alkaline conditions. Horseweed germination was > 20% at < 40 mM NaCl concentration and lowest (4%) at 160 mM NaCl. These data suggest that even at high soil salinity conditions, horseweed can germinate. Germination of horseweed decreased from 25% to 2% as osmotic potential increased from 0 (distilled water) to −0.8 MPa, indicating that germination can still occur under moderate water stress conditions. Horseweed seedling emergence was at its maximum on the soil surface, and no seedlings emerged from seeds placed at a depth of 0.5 cm or higher.

Nomenclature: Horseweed, *Conyza canadensis* (L.) Cronq. ERICA.

Key words: Emergence, light, osmotic stress, pH, photoperiod, salt stress, temperature.

Horseweed, also referred to as Canada fleabane or marestail, is a winter annual, native to North America (Bradshaw et al. 1997; Holm et al. 1997). An individual plant is capable of producing more than 200,000 seeds (Bhowmik and Bekech 1993; Weaver 2001). Seeds germinate best in early fall or spring; however germination can occur throughout the year (Buhler and Owen 1997; Holm et al. 1997; Regehr and Bazzaz 1979). Horseweed thrives in conservation- or no-tillage systems (Buhler 1992; Vencill and Banks 1994) but is susceptible to common tillage practices of conventional-tillage cropping systems (Brown and Whitwell 1988).

Increasing adoption of herbicide-resistant crop technology, especially glyphosate-resistant crops, has encouraged reduced and no-till production practices. This switch has encouraged traditionally noncropland weeds such as horseweed to move into and infest cropland. Compounding this problem, horseweed populations have developed resistance to glyphosate in several states, including Mississippi (Koger et al. 2004a). Populations of horseweed are also resistant to paraquat (Lehoczki et al. 1992; Smisek et al. 1998) and triazines (Lehoczki et al. 1984).

Various environmental components, such as temperature, light, pH, and soil moisture, have been known to influence weed seed germination (Chachalis and Reddy 2000; Koger et al. 2004b; Taylorson 1987). Little information exists about the effect of environmental factors on horseweed germination. Knowledge about horseweed germination would help in estimating the potential for its spread to new croplands. Therefore, the objective of this study was to determine the effect of temperature, light, pH, salt stress, and osmotic stress on horseweed germination.

Materials and Methods

Seed Source and General Experimental Procedure

Horseweed seeds were collected from naturally senescing plants in noncrop areas in Tunica County (34°42′N, 90°24′W) of Mississippi during late summer of 2002. Seeds were stored in screw-cap plastic bottles in the dark at 4 C until further use. Seeds used in all experiments, other than the temperature and light study, may have lost some viability due to a power failure in a storage facility for an undetermined amount of time. We have determined from preliminary experiments that 30 to 70 horseweed seeds per replication were adequate to measure effects on germination. Also, preliminary results have indicated no prechilling requirement for horseweed seeds, i.e., we did not find any dormancy in horseweed seed. On average, 50 seeds were evenly placed on a single layer of Whatman filter paper[1] in a 9-cm plastic petri dish. The filter paper was moistened with 5 ml of deionized water or treatment solution. This ensured uniform contact of treatment solutions with horseweed seeds. Petri dishes were sealed with parafilm[2] and then placed in black plastic trays. Trays were covered with aluminum foil for 24 h to acclimate seeds to growth conditions and to prevent the potential undesirable effects of exposure to bright light conditions, then trays were exposed to a photoperiod–temperature cycle of 13 h of light at 24 C and 11 h of dark at 20 C, unless specified otherwise. Germination measurements were taken at 7 and 10 d after treatment (DAT) in the temperature and light experiments, whereas all other experiments were terminated 7 DAT because no appreciable increase in horseweed germination occurred from 7 to 10 DAT. Seed was considered germinated when the radicle visibly protruded through the seed coat. Per-

centage of seed germination was calculated by multiplying the ratio of germinated seeds to total number of seeds in a single petri dish by 100.

Temperature and Light

Germination was determined in growth chambers under fluctuating temperatures (day/night temperatures: 36/30, 30/24, 24/20, 18/12, and 12/6 C). Temperature treatments were rotated between growth chambers for the second run of the experiments. Photoperiod was set at 13 h to coincide with the October day length. Fluorescent and incandescent lamps were used to produce a photosynthetic photon-flux density of 600 μmol m^{-2} s^{-1}. A set of petri dishes was wrapped in a layer of aluminum foil to study seed germination in the dark.

Rationale for Selection of Temperature and Light Regimes

Recent research in the field has shown that horseweed emerges throughout the year, with maximum emergence occurring in the fall (September to October) and a new flush emerging in the spring (April to May) (Koger et al. 2005). Thirty-year average weather data of Stoneville, MS, have indicated that the high temperature and low temperature in October and April (representing peak horseweed emergence periods in fall and spring, respectively) were 24 C and 11 C, respectively (Boykin et al. 1995). Temperature treatments were chosen to fall between 36 and 6 C to represent a broad range that overlapped the 11 to 24 C range. Also, the 30-yr average day length at Stoneville, MS, was 11.4 h in October and 13 h in April (Boykin et al. 1995). Hence, a photoperiod of 13 h was chosen to coincide with the maximum temperature in a temperature treatment, for example, 24 C in the 24/20 C treatment.

pH

The effect of pH on germination was studied using buffer solutions of pH 4 to 10 prepared as described for trumpetcreeper [*Campsis radicans* (L.) Seem. ex Bureau] seed (Chachalis and Reddy 2000). A 2-mM potassium hydrogen phthalate buffer solution was adjusted to pH 4 with 1 N HCl. A 2-mM solution of MES [2-(*N*-morpholino) ethanesulfonic acid] was adjusted to pH 5 and 6 with 1 N NaOH. A 2-mM solution of HEPES [*N*-(2-hydroxymethyl) piperazine-*N*-(2-ethanesulfonic acid)] was adjusted to pH 7 and 8 with 1 N NaOH. Buffer solutions of pH 9 and 10 were prepared with 2-mM tricine [*N*-Tris(hydroxymethyl) methylglycine] and adjusted with 1 N NaOH. Unbuffered deionized water (pH 6.7) was used as a control. Petri dishes were incubated at 24/20 C day/night temperature with 13 h light as described in the temperature and light study. These conditions provided the maximum germination in the temperature and light experiment.

Salt and Osmotic Stress

Sodium chloride solutions of 0, 10, 20, 40, 80, and 160 mM were prepared in deionized water. Petri dishes were incubated as described in the pH study. Aqueous solutions with osmotic potential of 0, −0.05, −0.10, −0.20, −0.40, and −0.80 MPa were prepared by dissolving appropriate amounts of polyethylene glycol (PEG) 8000 in deionized water (Steuter et al. 1981). Petri dishes were incubated as described in the pH study.

Planting Depth

Horseweed seeds were planted in soil[3] (Bosket sandy loam, fine-loamy, mixed, thermic Mollic Hapludalfs) in 10-cm-diam by 10-cm-deep plastic pots[4] at depths of 0, 0.2, 0.5, 1, 2.5, 5, and 7.5 cm. Pots were subirrigated as needed to maintain adequate soil moisture. Pots were covered with aluminum foil for 24 h and then exposed to a photoperiod-temperature cycle of 13 h of light at 24 C and 11 h of dark at 20 C. Germinated seedlings were counted 7 and 14 d after planting. Seedlings were considered emerged when the two cotyledons could be discerned visually.

Statistical Analysis

A completely randomized design with four replications was used in all experiments. Experiments were repeated. The data represent the average of the two experiments because of a nonsignificant experiment-by-treatment interaction and homogeneity of variance according to Bartlett's test, except for the pH experiments. The pattern of PH influence on horseweed germination was consistent between experiments; hence, data from a single experiment are reported. In all experiments, percentage of germination data were transformed using the $\log(x + 1)$ transformation, where x is percentage of germination. Transformation of data did not improve homogeneity; therefore, ANOVA and regression analysis were performed on nontransformed percentage of germination. Means from nonregressed experiments were separated using Fisher's protected LSD test at $P = 0.05$ or Duncan's new multiple range test at $P = 0.05$ (when the main treatment effect was not significant in ANOVA).

Results and Discussion

Temperature and Light

Temperature and light both significantly affected horseweed germination 7 and 10 DAT (Table 1). Horseweed seeds did not germinate when temperature was 12/6 C. As temperature increased, germination of horseweed seed increased and peaked at 24/20 C. Thereafter, germination decreased as temperature increased to 36/30 C. Horseweed seed germinated under both light (13-h photoperiod) and complete darkness (24 h), but the 13-h photoperiod was more favorable for germination than total darkness. These optimum temperature conditions for horseweed seed germination coincide with the typical late fall and early spring temperatures in the Mississippi Delta region. This light enhancement of seed germination has been reported in other weed species, including goosegrass [*Eleusine indica* (L.) Gaertn.] (Nishimoto and McCarty 1997), johnsongrass [*Sorghum halepense* (L.) Pers.] (Benech Arnold et al. 1990), and purple nutsedge (*Cyperus rotundus* L.) (Miles et al. 1996).

pH

Horseweed seed germination was 19 to 36% over a pH range from 4 to 10 (Figure 1). Seed germination was 24%

TABLE 1. Effect of temperature and light on horseweed germination.[a]

	Germination			
	Light (13 h photoperiod)		Dark (24 h)	
Temperature[b]	7 DAT	10 DAT	7 DAT	10 DAT
C	%			
12/6	0	0	0	0
18/12	15	32	7	12
24/20	60	61	14	15
30/24	22	—[c]	11	—
36/30	13	—	2	—
LSD (0.05)	21	7	9	7

	Germination		
	Light (13 h photoperiod)	Dark (24 h)	
	7 DAT	7 DAT	
Averaged across temperature regimes	25	7	
LSD (0.05)	8		

[a] Abbreviation: DAT, days after treatment.
[b] Maximum temperature within a regime corresponding to the 13 h photoperiod.
[c] Indicates that no further data were collected due to desiccation of seed under light and mold buildup under dark conditions.

or more when pH was 6 or higher. Horseweed seed germination was 19% at pH 4 or 5. These results suggest that horseweed tends to germinate better in neutral-to-alkaline soils compared with acidic soil environments. Similar to horseweed, other weed species, including common milkweed (*Asclepias syriaca* L.) (Evetts and Burnside 1972), redvine [*Brunnichia ovata* (Walt.) Shinners] (Shaw et al. 1991), sweet broomweed (*Scoparia dulcis* L.) (Jain and Singh 1989), texasweed [*Caperonia palustris* (L.) St. Hil.] (Koger et al. 2004b), and trumpetcreeper (Chachalis and Reddy 2000), germinated in a wide range of pH. In contrast, seeds of Canada thistle (*Cirsium arvense* L.) (Wilson 1979) and stranglervine (*Morrenia odorata* Lindl.) (Singh and Achhireddy 1984) germinated best between pH 6 to 7. Most Mississippi soils fall in the acidic-to-neutral range, whereas some soils have pH values greater than 7 (Anonymous 2003). Seed germination of horseweed over a broad range of pH has implications for widespread distribution and infestation in Mississippi.

Salt and Osmotic Stress

Germination of horseweed followed an exponential response to increasing salt concentration with germination decreasing as NaCl concentration increased from 20 to 160 mM (Figure 2). Horseweed germination was greater than 20% at < 40 mM NaCl concentration and lowest (4%) at 160 mM NaCl. These data suggest that horseweed may germinate at high soil salinity conditions. Chachalis and Reddy (2000) and Koger et al. (2004b) reported similar results for trumpetcreeper and texasweed, respectively.

A logarithmic equation best described germination of horseweed response to increasing osmotic potential (Figure 3). Germination of horseweed decreased from 25% to 2% as osmotic potential increased from 0 (distilled water) to −0.8 MPa. Although at lower incidence, horseweed can still

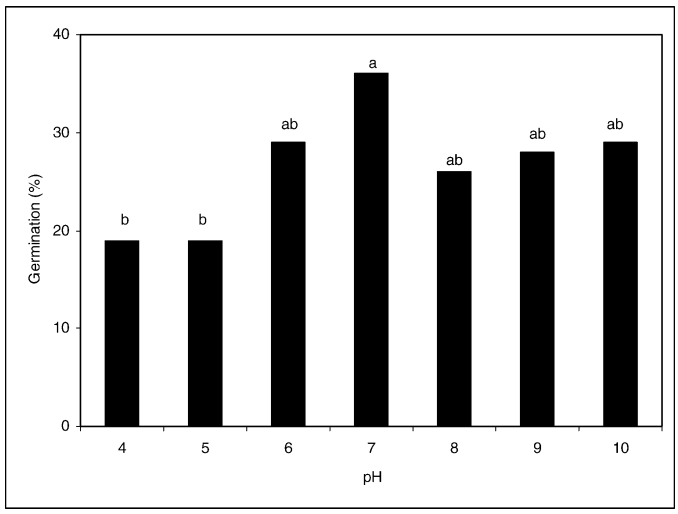

FIGURE 1. Effect of buffered pH solutions on germination of horseweed seed incubated at 24/20 C day/night temperature with a 13-h photoperiod for 1 wk. Bars with the same letters are not different (P = 0.05) according to Duncan's new multiple range test.

germinate under moderate water-stress conditions. Such conditions are typical during summer in the lower Mississippi River Delta region (Koger et al. 2004b). In contrast, germination of two other weed species commonly encountered in the southern United States, redvine (Shaw et al. 1991) and trumpetcreeper (Chachalis and Reddy 2000), was highly sensitive to low osmotic potential (< −0.2 MPa). Germination over a broad range of osmotic potential indicates that horseweed could pose a weed threat under both adequate and moisture-stress soil conditions.

Planting Depth

Horseweed emergence decreased dramatically with increasing planting depth (Table 2). Emergence was at its maximum for seeds planted on the soil surface, and no seedlings emerged from seeds placed at a depth of 0.5 cm or deeper. Bhowmik and Bekech (1993) reported 80% of ger-

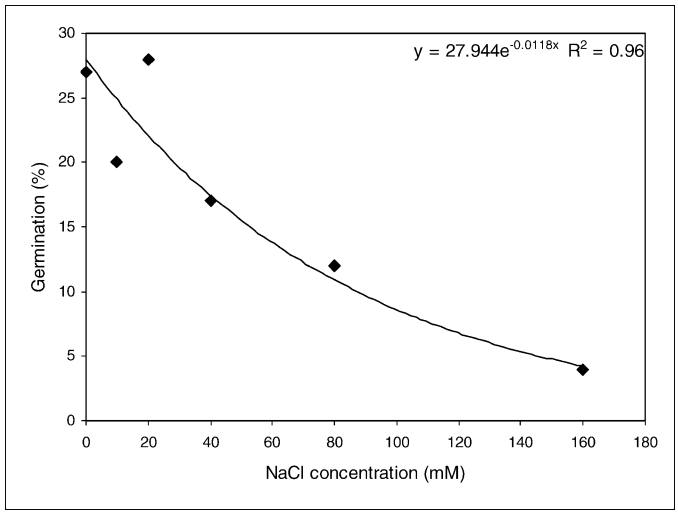

FIGURE 2. Effect of NaCl concentration on germination of horseweed seed incubated at 24/20 C day/night temperature with a 13-h photoperiod for 1 wk.

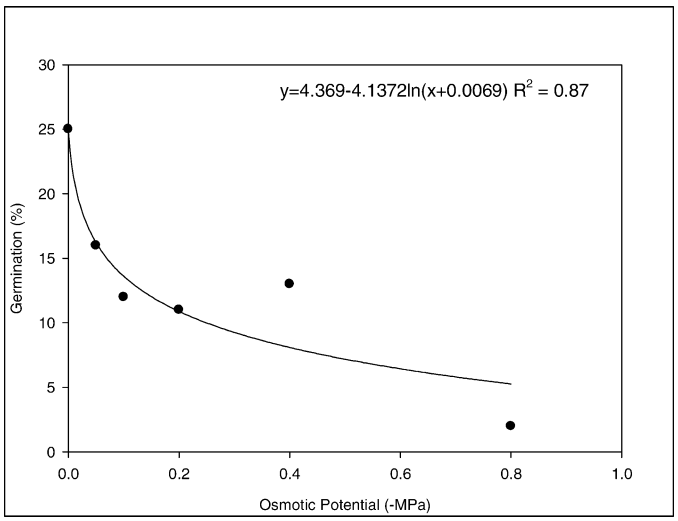

FIGURE 3. Effect of osmotic potential on germination of horseweed seed incubated at 24/20 C day/night temperature with a 13-h photoperiod for 1 wk.

minable horseweed seeds in the top 2 cm of soil from a no-tillage system, but no germinable seeds were found at depths greater than 6 cm. Horseweed emergence may also be affected by soil types. In the field, it has been noted that horseweed emerged faster and in greater numbers from coarse soils than from fine-textured soils (data not shown). VanGessel (2001) reported that horseweed emerged from a 0.5-cm depth in a commercial potting mixture, which was conceivably lighter than the soil type we used.

Emergence of weeds is affected by the amount of food reserves in seed and the depth of burial in the soil. Susceptibility of horseweed to common tillage practices associated with conventional cropping systems (Brown and Whitwell 1988) may be due to deep burial of extremely small horseweed seeds with limited reserves. Further, we have demonstrated that germination can be inhibited or delayed by darkness and enhanced by light. Seeds on the soil surface or buried shallowly can take advantage of light stimulation during germination and emergence. Decreased emergence due to increased planting depth has been reported in several weed species, including hairy beggarticks (*Bidens pilosa* L.) (Reddy and Singh 1992), horse purslane (*Trianthema portulacastrum* L.) (Balyan and Bhan 1986), and stranglervine (Singh and Achhireddy 1984). Under field conditions, no-tillage or minimal-tillage practices may encourage germination and emergence of horseweed.

In summary, these results suggest that horseweed has the ability to germinate under a broad range of environmental conditions. Prevailing conditions could determine the extent of germination, subsequent emergence, and overall severity of horseweed infestation.

Sources of Materials

[1] Whatman #1, Fisher Scientific, 711 Forbes Avenue, Pittsburgh, PA 15219.

[2] Parafilm, American National Can, American Lane, P.O. Box 3610, Greenwich, CT 06836.

[3] Research farm, Delta Research and Extension Center, Mississippi State University, Stoneville, MS 38776.

TABLE 2. Effect of planting depth on horseweed emergence.

Planting depth	Emergence	
	7 DAP[a]	14 DAP
cm	# seedlings pot^{-1}	
0	100	113
0.25	3	5
0.5	0	0
1	0	0
2.5	0	0
5	0	0
7.5	0	0
LSD (0.05)	6	6

[a] Abbreviation: DAP, days after planting.

[4] National Polymers Inc., 7920 West 215th Street, Lakeville, MN 55044.

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Received January 5, 2006, and approved June 5, 2006.